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**ABSTRACT:** Present report focuses on the morphometrical and morphological study of SEM of endoparasites exclusively of metacercaria (the encysted form) of trematode parasite found in the liver of most edible fish Channa punctatus of Moradabad region. In our investigation the liver of host was found highly infected with eggs of trematode and unidentified DTM which is characterized by a complicated morphology of its integument, body and suckers. The topography of egg was characterized by 2.34 mm × 1.17 mm whereas entire body of DTM was observed 3.38 mm × 1.50 mm in diameter having two sophisticated suckers to absorb the nutrients from host body and integument of metacercaria was found traversed with numerous sensory papillae which may be helpful in taxonomical study of parasite. Present findings underscore the necessity of awareness to prevent endemic effects due to fish borne zoonotic trematode (FZT) causing increased risk to public health.

**Index Terms:** morphology, endoparasite, digenean trematode metacercarieae (DTM), fish borne zoonotic trematode (FZT).

1. INTRODUCTION
Helminthes are common parasites found in wild and cultured fish. A host may be infected simultaneously one, two or all varieties of parasites mainly nematodes cestodes, and trematodes. Digeneric trematodes are the parasites of several hosts such as snails, fishes and birds. Many fishes are the host of these parasites which are varying in shape and size with a complex life cycle. Hussein et al (2010) studied the ultra structure of eggs and larval stages including redia, cercaria and metacercaria of trematode Fasciola gigantica, from infected Lymnaea (cailliaudi) natalensis snails using light and Scanning Electron Microscopy. Many fishes have been reported as the intermediate hosts of Clinostomum spp. (Aohagi et al 1993), however Yamaguti (1971) categorised taxonomically various digeneric trematode of vertebrates. Chai et al (2017) investigated infestation status of metacercaria trematode in Yangoon and Myanmar fishes. Moema et al (2013) performed SEM study for ultrastructure of two diplostomids parasites found in the Clarias gariepinus. Raimundo et al. (2014) reported two species of trematode metacercaria, Clinostomatopsis sorbens and Ithyoclinostomum dimorphum in Hoplyertrinus unliaenatis (gold wolf fish) of Brazil. Tondon and Roy (2002) observed some peculiarities of the metacercaria like its small, elongated body, and presence of oral sucker and acetabulum Barson et al (2008) reported the morphological measurements of the helmith parasites found from Clarias gariepinus. Metacercaria are generally found embedded in the host liver therefore destruction of liver cells caused abundantly, these destructive structures called as lesions which usually forms on the attachment site of the host were observed by Goselle et al. (2008). Palm and Overstreet (2000) observed that trematode were embedded in liver wall of host and covered with cyst and they suggested that parasites might be transferred from fishes to humans (zooneses). Metacercariae characteristics were also described by Kanev et al. (2002). Swarnakar et al (2014) in their SEM study reported two types of suckers placed anterior and ventral region of the trematode Orthocoeoelium scolocoeoelium recovered from buffalo. The integument is the interfacing layer that helps the parasite to maintain homeostasis and evade hostile environments including the host’s immune attacks and also a suitable place for multitudes of papillae, which were detected on the outer surface of metacercaria as reported by Marwan and Mohammed (2003). Two types of the structures including spines and sensory papillae and structural variations in both were observed by Nahed and Ashour (2000) in their work on SEM of tegument surface of digeneric trematode Stephanostomum egypticum from the red sea fishes. Sohn et al (2016) suggested that large scale findings of these trematodes play the key role in the human infection. Eiras et al (2010) observed in their findings that zoonotic illness is being caused by metacercaria in Brazil. Keiser and Utzinger (2005) reported considerable risk to public health due to trematode fish parasites. These metacercariae directly affect the fish eaters if fish is being eaten in unhygienic way.

2. MATERIALS AND METHODS
The fish samples of Channa punctatus were obtained from various water bodies of Moradabad i.e. river Ramganga, and its tributaries. Trematode parasites were collected from freshly killed samples from the host liver and the body cavity. The encysted metacercariae of trematode were separated into saline solution to remove their cyst. Excysted metacercariae were fixed in 2.5 normal gluteraldehyde for 4 hrs, washed in 01% buffer solution and post fixed in osmium tetraoxide solution (Bancroft and Stevens, 1977). After removal from osmium tetraoxide metacercariae were treated with the graded alcohol series followed by gold coating chamber then examined the ultra structure of the parasite by the help of JEOL JSM 649 scanning electron microscope.

3. RESULT AND DISCUSSION
Fig.1 represents the encysted metacercariae collected from liver separated for further SEM study. The metacercariae are fine miniature having sharp and oval body shape with non spiny outer surface. Parasites were directly connected to the fish liver by the help of its tegument (fig.1a). Egg of trematode was small, smooth and oval shaped, pale yellow in colour with flabby appearance of cytoplasmic bulging on outer surface (fig 1b) and was measuring about 2.34mm × 1.17 mm in length and width on the magnification of 37X.
Hussein et al (2010) also reported smooth outer surface of the egg shell having no micro spines in egg and metacercaria of trematode Fasciola species as similar with our findings of smooth egg surface and absence of any microspines on the egg of trematode metacercaria isolated from the liver of C. punctatus. The whole body of metacercaria on the magnification of 27X was observed as 3.3mm × 1.10mm in length and width to the anterior extremity while 1.50 mm width from the posterior extremity (fig 1c). The ultra structure of excysted metacercaria shows oral sucker and ventral sucker at X120 and X100 magnification respectively (fig 1c and 1d) and the tip of the oral sucker was devoid of any tegumental spine (fig. 2b). On the anterior side of body protrusion oral sucker was found slightly perforated in the centre which may support in penetration to the host body. Ventral sucker observed just below the wrinkles (fig. 1d and 1e) measures about 491.63μm × 508.02μm in diameter from outside and 280.11μm in length from inside at X100 is also known as the acetabulum i.e. a circular depression which helps in attachment placed ventrally along side of the mouth. Morsy et al (2018) in their SEM study reported the oral sucker contained a terminal mouth which is longer than its width and the lip of ventral sucker was covered by wrinkles of integument in digenean parasite Urotrema scabridum which were quite similar to our findings of shape and position of suckers in metacercaria (fig. 1d) isolated from C. punctatus. A lot of sensory papillae were present (fig 2b) over the outer surface of integument of metacercaria including suckers and also found traversed with the cytoplasmic ridges and furrows as detected by scanning topography in our report however Chai et al (2016) reported the arrangement of small sensory papillae around the oral sucker and large papillae around the ventral sucker of the parasite Stellantchasmus falcatus. Morphological differentiation of finger like protrusions in tegument of metacercaria were also reported by Nahed and Ashour (2000) in Stephanostomum egypticum recovered from the red sea fishes similar as our findings of the tegument in metacercaria. The tegumental modifications in metacercariae were also observed by Sethukarasi and Veerakumari (2007) on the surface topography of a few digeneans (Family: Didymozoidae). In our findings metacercaria was found from liver of the host, whereas Aziza and Torkia (2003) recovered metacercaria mainly from the tissues around gills and rarely from the body cavity, in their work on SEM of Clinostomum metacercaria isolated from Oreochromis niloticus from Assiut, Egypt. In the present study the encysted metacercaria were found as their cyst is supposed to be a protective layer for larvae against the host enzyme. Similar type of findings in trematode metacercaria were also described earlier by Hung et al (2013) whereas Choi et al (1995) reported naked metacercaria isolated from oyster Crassostrea gigas. (fig. 1b). Similar type of findings in trematode metacercaria were also described earlier by Hung et al (2013) whereas Choi et al (1995) reported naked metacercaria isolated from oyster Crassostrea gigas.
Fig. 2. a: oral and ventral suckers on X85 (magnification), b: cytoplasmic bulging and papillae on the outer surface X300.

Usually the difference in specific structures of all trematode metacercaria has been reported in various SEM studies by different workers as cited above may be due to its presence to the site of occurrence, homeostasis, immune-response relationship of parasite-host biological system and many other unknown factors still to be categorized.

4. Conclusion

The edible fish C. punctatus of Ramganga found infected with metacercaria of trematode parasite is a causative agent of zoonotic disease. The SEM study provides a specific characteristics and ultra-structure of these parasites which may be helpful to find and identify the various types of trematode parasites which are harmful to fish-eaters.

References

[18] H.W, Overstreet, Ototobium cysticum (Cestoda: Trypanorhynchia) from the muscle of butterfishes.


